

CLAIMS

1. An isolated oligonucleotide that

(a) comprises a nucleotide sequence that is complementary to
5 a region consisting of:

(i) the central repeat unit of three repeat units composing
a tandem repeat in the promoter region of the thymidylate
synthase gene, and

10 (ii) the repeat unit located downstream of the central
repeat unit,

and (b) hybridizes to the region of (a) under highly stringent
hybridization conditions.

2. The isolated polynucleotide of claim 1, wherein the
nucleotide hybridizes to the 3' end repeat unit of the two
15 repeat units composing a tandem repeat in the promoter region
of the thymidylate synthase gene, under hybridization
conditions that are less stringent than (b).

3. The isolated oligonucleotide of claim 2, wherein the
nucleotide sequence comprises the nucleotide sequence of SEQ
20 ID NO: 2.

4. An isolated oligonucleotide that hybridizes to the region
adjacent to 5' side of the oligonucleotide that:

(a) comprises a nucleotide sequence that is complementary
to a region consisting of:

25 (i) the central repeat unit of three repeat units composing
a tandem repeat in the promoter region of the thymidylate
synthase gene, and

(ii) the repeat unit located downstream of the central
repeat unit, and

30 (b) hybridizes to the region of (a).

5. The isolated oligonucleotide of claim 4, wherein the
nucleotide sequence comprises the nucleotide sequence of SEQ
ID NO: 1.

6. A method for identifying the number of tandem repeats in the
35 promoter region of the thymidylate synthase gene, the method
comprising:

(a) amplifying a genomic DNA that comprises tandem repeats in at least the promoter region of the thymidylate synthase gene,

(b) hybridizing the oligonucleotide of claim 1 to the amplified genomic DNA of step (a) under stringent conditions,

(c) detecting a hybridization between the oligonucleotide and the genomic DNA, and

(d) identifying the number of tandem repeats as "two" when hybridization is not detected, identifying the number of tandem repeats as "three" when hybridization is detected,

7. The method of claim 6, further comprising:

(e) hybridization the oligonucleotide of claim 1 to the amplified genomic DNA of step (a) under hybridization conditions that are less stringent than (b),

(f) detecting a hybridization between the oligonucleotide and the genomic DNA, and

(g) identifying the number of tandem repeats as "two" when hybridization is not detected in (c) but is detected in (f),

8. The method of claim 6, wherein the hybridization is detected by melting curve analysis.

9. The method of claim 8, the method comprising the step of detecting fluorescence resonance energy transfer using (i) the oligonucleotide of claim 4, wherein the 3' end of the oligonucleotide is labeled with a fluorescent dye, and (ii) the oligonucleotide of claim 1 whose 5' end is labeled with a different fluorescent dye that transfers fluorescence resonance energy to the fluorescent dye at the 3' end of the oligonucleotide of (i).

10. The method of claim 9, wherein the oligonucleotide of (ii) comprises the nucleotide sequence of SEQ ID NO: 2.

11. The method of claim 10, wherein the oligonucleotide of (i) comprises the nucleotide sequence of SEQ ID NO: 1.

12. The method of claim 9, wherein the fluorescent dye that labels the oligonucleotide of (i) is FITC, and the fluorescent dye that labels the oligonucleotide of (ii) is RED640 or RED705.

13. A method for genotyping the thymidylate synthase gene of a subject, the method comprising:

(a) identifying the number of tandem repeats in the promoter region of the thymidylate synthase gene by the method of claim 5 6, and

(b) determining that the thymidylate synthase genotype of the subject is "homozygous 2R/2R" when the number of tandem repeats is identified as only two, "homozygous 3R/3R" when the number of tandem repeats is identified as only three, 10 or "heterozygous 2R/3R" when the number of tandem repeats is identified as both "two" and "three".

14. A method for predicting the responsiveness of a subject towards an antitumor agent targeting thymidylate synthase, the method comprising:

15 (a) determining the thymidylate synthase genotype of the subject by the method of claim 13, and

(b) associating the thymidylate synthase genotype with the responsiveness of the subject towards an antitumor agent targeting thymidylate synthase.

20 15. A method for determining the dose and/or the type of an antitumor agent targeting thymidylate synthase for treating a cancer patient, the method comprising:

(a) determining the thymidylate synthase genotype of the patient by the method of claim 13, and

25 (b) for a "homozygous 2R/2R" patient, deciding to: (i) administer an antitumor agent dose that is lower than the normally used dose, or (ii) use an antitumor agent that has a different target.

30 16. A kit for identifying the number of tandem repeats in the promoter region of the thymidylate synthase gene, the kit comprising:

(i) the oligonucleotide of claim 4, and

(ii) the oligonucleotide of claim 1.

35 17. The kit of claim 16, wherein 3' end of the oligonucleotide of (i) is labeled with FITC, and the 5' end of the oligonucleotide of (ii) is labeled with the fluorescent dye.

RED640 or RED705.

18. The kit of claim 16, the kit comprising:

- (a) an oligonucleotide comprising the nucleotide sequence of SEQ ID NO: 1, and
- 5 (b) an oligonucleotide comprising the nucleotide sequence of SEQ ID NO: 2.